Wrapup 2008-06-10

Priority

General

Plasmids

pLV-TRE-Ngn1-EYFP-2A-mKate-Ubc-Bla

[1] PCR SOEing: Ngn1-EYFP-2A-mKate – Started PCR amplify for Ngn1, EYFP, mKate

[1] Redo PCR for Ngn1-EYFP

[1] Make gel

[1] Run gel of PCR products: Ngn1, EYFP, mKate
– Ngn1: didn't do. Need to wait for correct primer
-- EYFP: band around 1kb ??
-- mKate: two bands

[1] Extract PCR products from gel
-- cut out gel for EYFP and mKate

[2] Parent vector: pLV-TRE-Sox17-Ubc-Bla

[2] Pick colonies of pLV-TRE-Sox17-Ubc-Bla (10 total, 5 from each of two plates)

[2] Grow minipreps – Prepare growth cultures; 12

pLV-Ubc-rtTA-2A-Bla

[2] Ligate pFUGW-AgeI-EcoRI-CIP and rtTA-2A-Bla-BspEI-EcoRI

[2] Transformation of ligation. Grow overnight on plates (Andrew).

p148

[3] Transform p148. Grow overnight on plates (Evan).

p149

[3] Transformed

[3] Pick colony

[3] Grow colony for maxiprep. Take out into +4C Wed. @ 8:30am (Navin)

[3] Maxiprep

[3] Restriction mapping (plasmid?)

Lentivirus

Lenti: pLV-TRE-Ngn1-EYFP-2A-mKate-Ubc-Bla

Experiments

Experiment: Ngn1-EYFP-2A-mKate

Infect cells

Add Dox

Observe differentiation

Make Yellow/Red artificial brains